

Published in "European Journal of Clinical Microbiology & Infectious Diseases
doi: 10.1007/s10096-017-3063-z, 2017"
which should be cited to refer to this work.

Activity of the novel siderophore cephalosporin cefiderocol against multidrug-resistant Gram-negative pathogens

J. Dobias^{1,2,3,4} • V. Dénervaud-Tendon^{1,2,3,4} • L. Poirel^{1,2,3,4} • P. Nordmann^{1,2,3,4,5}

Abstract The novel siderophore cephalosporin cefiderocol (S-649266) with potent activity against Gram-negative pathogens was recently developed (Shionogi & Co., Ltd.). Here, we evaluated the activity of this new molecule and comparators against a collection of previously characterized Gram-negative isolates using broth microdilution panels. A total of 753 clinical multidrug-resistant Gram-negative isolates collected from hospitals worldwide were tested against cefiderocol and antibiotic comparators (ceftolozane–tazobactam [CT], meropenem [MEM], ceftazidime [CAZ], ceftazidime–avibactam [CZA], colistin [CST], aztreonam [ATM], amikacin [AMK], ciprofloxacin [CIP], cefepime [FEP], and tigecycline [TGC]) for their susceptibility. The collection included *Escherichia coli* ($n = 164$), *Klebsiella pneumoniae* ($n = 298$), *Enterobacter* sp. ($n = 159$), *Pseudomonas aeruginosa* ($n = 45$), and *Acinetobacter baumannii* ($n = 87$). Resistance mechanisms included producers of carbapenemases and extended-spectrum β -lactamases (ESBLs). In addition, a series of colistin-resistant enterobacterial isolates ($n = 74$), including 15 MCR-1 producers, were tested. The MIC₉₀ of cefiderocol was 2 mg/

L, while those of comparative drugs were >64 mg/L for CT, MEM, CAZ, CZA, and AMK, >32 mg/L for ATM, >16 mg/L for FEP, 8 mg/L for CST, and 2 mg/L for TGC. The MIC₅₀ of cefiderocol was 0.5 mg/L, while those of other drugs were >64 mg/L for CAZ, 64 mg/L for CT, >32 mg/L for ATM, >16 mg/L for FEP, 8 mg/L for MEM and AMK, >4 mg/L for CIP, 1 mg/L for CZA, 0.5 mg/L for TGC, and <0.5 mg/L for CST. Only 20 out of 753 strains showed MIC values of cefiderocol ≥ 8 $\mu\text{g/mL}$. Compared to the other drugs tested, cefiderocol was more active, with the exception of colistin and tigecycline showing equivalent activity against certain subgroups of bacteria.

Introduction

Gram-negative bacteria that produce extended-spectrum β -lactamases (ESBLs) are a major concern in healthcare due to their ability to spread globally [1]. ESBLs are a major group of enzymes that confer resistance to several generations of β -lactam antibiotics, including third-generation cephalosporins [2, 3]. ESBL-encoding genes that are primarily plasmid-encoded include mostly TEM-, SHV-, and CTX-M-type enzymes [4]. Enterobacteriaceae, such as *Klebsiella pneumoniae* and *Escherichia coli*, are the main ESBL producers that have been reported globally. Carbapenems are mostly used for the treatment of infections due to ESBL-producing bacteria, but carbapenemase-producing bacteria are now extensively reported [5, 6]. Polymyxins (e.g., colistin and polymyxin B) are now more frequently used as last-resort antibiotics for treating patients with multidrug-resistant bacterial infections [7]. However, a recent report shows that a novel gene (*mcr-I*) as a source of plasmid-encoded resistance encoded on a plasmid in *E. coli* may quickly spread to other bacterial strains or species [8–10]. Therefore, due to the increasing threat of

✉ P. Nordmann
patrice.nordmann@unifr.ch

¹ Emerging Antibiotic Resistance Unit, University of Fribourg, Fribourg, Switzerland

² National Reference Center for Emerging Antibiotic Resistance (Switzerland), University of Fribourg, Fribourg, Switzerland

³ French INSERM European Unit (LEA/IAME), University of Fribourg, Fribourg, Switzerland

⁴ Medical and Molecular Microbiology Unit, Department of Medicine, University of Fribourg, rue Albert Gockel 3, 1700 Fribourg, Switzerland

⁵ Institute for Microbiology, University of Lausanne and University Hospital Centre, Lausanne, Switzerland

multidrug-resistant and pandrug-resistant bacteria, there is a need for novel molecules. Cefiderocol (CFDC) is a novel parenteral siderophore cephalosporin also known as S-649266. It possesses a unique mechanism for penetrating efficiently into Gram-negative pathogens. It uses a “Trojan horse” strategy by binding free iron and is then actively transported into bacterial cells across the outer membrane of Gram-negative bacteria by way of the iron-transport system [11, 12]. Cefiderocol is a cephalosporin molecule with an attached catechol moiety on the 3-position side chain which binds to ferric iron [11]. Once across the outer membrane, the iron dissociates and the cephalosporin binds to penicillin-binding proteins (PBP), mainly PBP3, as other cephalosporins do to disrupt cell wall synthesis [11, 13], contributing to a potent antimicrobial activity against Gram-negative bacteria. In addition, this antimicrobial activity of cefiderocol is enhanced by the high stability of cefiderocol to hydrolysis by nearly all β -lactamases, including both the serine and metallo-carbapenemases [14]. The ability to cross the outer membrane through the active iron-transport system overcomes resistance due to porin channel mutations and efflux pump overproducers. It results that cefiderocol exhibits potent in vitro and in vivo activity against all species of Gram-negative bacteria, including carbapenem-resistant strains of Enterobacteriaceae, *Pseudomonas aeruginosa*, and *Acinetobacter baumannii*, and even *Stenotrophomonas maltophilia* [15]. Here, we evaluated the antimicrobial activity of cefiderocol and other Gram-negative antibiotics (aztreonam, amikacin, cefepime, ceftazidime, ceftazidime–avibactam, ceftolozane–tazobactam, ciprofloxacin, meropenem, colistin, and tigecycline) against a panel of 753 multidrug-resistant bacterial isolates from human clinical sources with characterized antibiotic resistance mechanisms.

Materials and methods

Bacterial isolates

A total of 753 clinical multidrug-resistant isolates were evaluated in this study (Table 1). They were representative of the most widespread and broad-spectrum mechanisms of resistance currently observed worldwide in Gram-negative bacteria. The strains were collected from hospitals worldwide (42 countries) from 2000 to 2016, with a majority dating from the 2012–2016 period. They were of various origins (not always recorded) but mostly from urines, broncho-alveolar specimens, blood, pus, and stools.

In vitro susceptibility test methods

Minimum inhibitory concentrations (MICs) were determined following the Clinical and Laboratory Standards Institute (CLSI) broth microdilution (BMD) guidelines [16]. Frozen

Table 1 Bacterial strains tested in this study

Genus (species)	Number of tested isolates	Characterized resistance
<i>Escherichia coli</i> (164)		
	43	OXA (-1/48/181/204)
	22	VIM (-1/2/4/19), IMP (-1/8)
	45	NDM (-1/4/5/6/7)
	11	KPC (-2/3)
	25	CTX-M (-1/3/15)
	15	MCR-1
	3	Non-MCR-1 colistin resistant
<i>Klebsiella pneumoniae</i> (298)		
	101	KPC (-2/3/11)
	89	OXA (-48/162/163/181/204/232) ^a
	18	NDM ^b (-1/4)
	20	VIM (-1/4/19), IMP (-1/4/8)
	25	CTX-M (-3/15)
	45	Colistin R
Enterobacteriaceae (159)		
	26	OXA (-48/163)
	10	NDM-1
	5	VIM-1
	14	IMP (-1/8)
	14	KPC-2
	49	CTX-M (-3/15)
	7	VEB-1
	14	SHV-12
	9	Plasmid-mediated AmpC and overproducer of AmpC
	11	Colistin R
<i>Pseudomonas aeruginosa</i> (45)		
	6	PER-1
	9	SHV (-2a/5/12), GES (-1/9)
	20	IMP (-1/2/10/13/15/19/29), KPC-2
	10	VIM (-1/2), SPM-1, GIM-1
<i>Acinetobacter baumannii</i> (87)		
	85	OXA-23/40/58/72
	2	NDM-1, IMP-4
Total	753	

^a Five strains are OXA-48 producers and colistin resistant

^b Five strains are NDM producers and colistin resistant

96-well broth microdilution panels with pre-loaded antibiotic-growth medium were supplied by International Health Management Associates, Inc. (Schaumburg, IL, USA). Cefiderocol was tested in iron-depleted cation-adjusted Mueller–Hinton broth (ID-CAMHB) [17], as recently approved by the CLSI (<http://clsi.org/standards/micro/microbiology-files/>), whereas comparators were tested in cation-adjusted Mueller–Hinton broth (CAMHB). Quality

Table 2 In vitro activities of cefiderocol (CFDC), ceftolozane-tazobactam (CT), meropenem (MEM), ceftazidime (CAZ), ceftazidime-avibactam (CZA), colistin (CST), aztreonam (ATM), amikacin (AMK), ciprofloxacin (CIP), cefepime (FEP), and tigecycline (TGC) against *E. coli*, *K. pneumoniae*, *Enterobacter* sp., *P. aeruginosa*, and *A. baumannii*

Organism group/ antimicrobial agent	MIC range (mg/L)	No.	MIC ₅₀ ^a	MIC ₉₀ ^a
<i>All species</i>				
CFDC	0.03–64	753	0.5	2
CT	0.03–64	753	64	>64
CAZ	0.03–64	753	>64	>64
CZA	0.03–64	753	1	>64
FEP	0.05–16	753	>16	>16
MEM	0.03–64	753	8	>64
CIP	0.25–4	753	>4	>4
ATM	0.05–32	753	>32	>32
AMK	4–64	753	8	>64
CST	0.5–8	753	≤0.5	8
TGC	0.25–4	753	0.5	2
<i>Escherichia coli</i>				
CFDC	0.03–64	164	0.5	4
CT	0.03–64	164	8	>64
CAZ	0.03–64	164	64	>64
CZA	0.03–64	164	0.5	>64
FEP	0.05–16	164	>16	>16
MEM	0.03–64	164	2	>64
CIP	0.25–4	164	>4	>4
ATM	0.05–32	164	>32	>32
AMK	4–64	164	≤4	>64
CST	0.5–8	164	≤0.5	2
TGC	0.25–4	164	≤0.25	≤0.25
<i>Klebsiella pneumoniae</i>				
CFDC	0.03–64	298	1	2
CT	0.03–64	298	64	>64
CAZ	0.03–64	298	>64	>64
CZA	0.03–64	298	1	>64
FEP	0.05–16	298	>16	>16
MEM	0.03–64	298	16	>64
CIP	0.25–4	298	>4	>4
ATM	0.05–32	298	>32	>32
AMK	4–64	298	8	64
CST	0.5–8	298	≤0.5	16
TGC	0.25–4	298	0.5	1
<i>Enterobacter</i> sp.				
CFDC	0.03–64	159	0.5	4
CT	0.03–64	159	16	>64
CAZ	0.03–64	159	>64	>64
CZA	0.03–64	159	0.5	>64
FEP	0.05–16	159	>16	>16
MEM	0.03–64	159	0.5	16
CIP	0.25–4	159	>4	>4
ATM	0.05–32	159	>32	>32
AMK	4–64	159	≤4	16
CST	0.5–8	159	≤0.5	8
TGC	0.25–4	159	0.5	1

Table 2 (continued)

Organism group/ antimicrobial agent	MIC range (mg/L)	No.	MIC ₅₀ ^a	MIC ₉₀ ^a
<i>Pseudomonas aeruginosa</i>				
CFDC	0.03–64	45	0.5	2
CT	0.03–64	45	64	>64
CAZ	0.03–64	45	>64	>64
CZA	0.03–64	45	16	>64
FEP	0.05–16	45	>16	>16
MEM	0.03–64	45	32	>64
CIP	0.25–4	45	>4	>4
ATM	0.05–32	45	16	>32
AMK	4–64	45	8	>64
CST	0.5–8	45	≤0.5	1
TGC	0.25–4	45	>4	>4
<i>Acinetobacter baumannii</i>				
CFDC	0.03–64	87	0.12	4
CT	0.03–64	87	16	>64
CAZ	0.03–64	87	>64	>64
CZA	0.03–64	87	16	64
FEP	0.05–16	87	>16	>16
MEM	0.03–64	87	32	64
CIP	0.25–4	87	>4	>4
ATM	0.05–32	87	>32	>32
AMK	4–64	87	64	>64
CST	0.5–8	87	≤0.5	1
TGC	0.25–4	87	1	2

^a MIC, minimum inhibitory concentration; MIC_{50/90}, MIC that inhibits 50% and 90% of the isolates, respectively

control (QC) strains (*E. coli* ATCC 25922, *K. pneumoniae* ATCC 700603, and *P. aeruginosa* ATCC 27853) were tested regularly to ensure the stability of the panels and validity of the test methods. The acceptable concentration range for QC strains is 0.06–0.5 mg/L for *E. coli* and *P. aeruginosa* and not yet defined for *K. pneumoniae*.

All isolates were tested against the following antibiotics for the given concentration range: cefiderocol (S-649266) (CFDC, 0.03–64 mg/L), aztreonam (ATM, 0.5–32 mg/L), ceftazidime (FEP, 0.5–16 mg/L), ceftolozane–tazobactam (CT, 0.03–64 mg/L), meropenem (MEM, 0.03–64 mg/L), ceftazidime (CAZ, 0.03–64 mg/L), ceftazidime–avibactam (CZA, 0.03–64 mg/L), colistin (CST, 0.5–8 mg/L), amikacin (AMK, 4–64 mg/L), ciprofloxacin (CIP, 0.25–4 mg/L), and tigecycline (TGC, 0.25–4 mg/L).

The strains were grown overnight on a non-selective agar medium and two to three colonies were solubilized in 3 mL sterile 0.85% NaCl. The turbidity was adjusted to 0.5 McFarland with a McFarland densitometer DEN-1B from Grant Bio (Grant instruments Ltd., Cambridge, UK). One milliliter of inoculum was then diluted in 29 mL of sterile ddH₂O and 10 µL were added to each BMD panel well. The panels were incubated for 16–20 h at 35 °C. To ensure an even

thermal distribution during incubation, the panels were stacked no more than four high. The MIC reading was then done according to the CLSI guidelines [18].

Results and discussion

The susceptibility test results are listed in Tables 2–7. The MIC₉₀ (MIC value that inhibits 90% of the isolates) of cefiderocol was 2 mg/L (Table 2), while those of comparative drugs were >64 mg/L for CT, MEM, CAZ, CZA, and AMK, >32 mg/L for ATM, >16 mg/L for FEP, 8 mg/L for CST, and 2 mg/L for TGC. The MIC₅₀ of CFDC was at 0.5 mg/L, while those of other drugs were >64 mg/L for CAZ, 64 mg/L for CT, >32 mg/L for ATM, >16 mg/L for FEP, 8 mg/L for MEM and AMK, >4 mg/L for CIP, 1 mg/L for CZA, 0.5 mg/L for TGC, and ≤0.5 mg/L for CST. The addition of 4 µg/mL of avibactam restored the activities of ceftazidime against the majority of the tested enterobacterial isolates.

The MIC₅₀ and MIC₉₀ of CFDC for Enterobacteriaceae producing KPC carbapenemases were ≤1 µg/mL and ≤2 mg/L, respectively (Table 3). The only competitive comparators were ceftazidime–avibactam (MIC_{50/90}) (1/4), colistin (≤0.5/

Table 3 In vitro activities of cefiderocol (CFDC), ceftolozane-tazobactam (CT), ceftazidime (CAZ), ceftazidime-avibactam (CZA), cefepime (FEP), meropenem (MEM), ciprofloxacin (CIP), aztreonam (ATM), amikacin (AMK), colistin (CST), and tigecycline (TGC) against Enterobacteriaceae (*E. coli*, *K. pneumoniae*, and *Enterobacter* sp.) producing a KPC-type carbapenemase

Organism group/antimicrobial agent	MIC range (mg/L)	No.	MIC ₅₀ ^a	MIC ₉₀ ^a
<i>Total Enterobacteriaceae</i>				
CFDC	0.03–64	127	1	2
CT	0.03–64	127	64	>64
CAZ	0.03–64	127	>64	>64
CZA	0.03–64	127	1	4
FEP	0.05–16	127	>16	>16
MEM	0.03–64	127	32	>64
CIP	0.25–4	127	>4	>4
ATM	0.05–32	127	>32	>32
AMK	4–64	127	16	32
CST	0.5–8	127	≤0.5	>8
TGC	0.25–4	127	0.5	1
<i>Escherichia coli</i>				
CFDC	0.03–64	12	0.5	1
CT	0.03–64	12	32	>64
CAZ	0.03–64	12	64	>64
CZA	0.03–64	12	1	4
FEP	0.05–16	12	>16	>16
MEM	0.03–64	12	8	64
CIP	0.25–4	12	>4	>4
ATM	0.05–32	12	64	64
AMK	4–64	12	≤4	>64
CST	0.5–8	12	≤0.5	≤0.5
TGC	0.25–4	12	≤0.25	≤0.25
<i>Klebsiella pneumoniae</i>				
CFDC	0.03–64	101	1	2
CT	0.03–64	101	64	>64
CAZ	0.03–64	101	>64	>64
CZA	0.03–64	101	2	4
FEP	0.05–16	101	>16	>16
MEM	0.03–64	101	64	>64
CIP	0.25–4	101	>4	>4
ATM	0.05–32	101	>32	>32
AMK	4–64	101	16	32
CST	0.5–8	101	≤0.5	>8
TGC	0.25–4	101	0.5	1
<i>Enterobacter</i> sp.				
CFDC	0.03–64	14	0.5	1
CT	0.03–64	14	64	>64
CAZ	0.03–64	14	>64	>64
CZA	0.03–64	14	1	2
FEP	0.05–16	14	>16	>16
MEM	0.03–64	14	8	16
CIP	0.25–4	14	>4	>4
ATM	0.05–32	14	>32	>32
AMK	4–64	14	≤4	≤4
CST	0.5–8	14	≤0.5	8
TGC	0.25–4	14	0.5	1

^a MIC, minimum inhibitory concentration; MIC_{50/90}, MIC that inhibits 50% and 90% of the isolates, respectively

Table 4 In vitro activities of cefiderocol (CFDC), ceftolozane-tazobactam (CT), ceftazidime (CAZ), ceftazidime-avibactam (CZA), cefepime (FEP), meropenem (MEM), ciprofloxacin (CIP), aztreonam (ATM), amikacin (AMK), colistin (CST), and tigecycline (TGC) against Enterobacteriaceae (*E. coli*, *K. pneumoniae*, and *Enterobacter* sp.) producing an OXA-48-type carbapenemase

Organism group/antimicrobial agent	MIC range (mg/L)	No.	MIC ₅₀ ^a	MIC ₉₀ ^a
<i>Total Enterobacteriaceae</i>				
CFDC	0.03–64	154	0.25	2
CT	0.03–64	154	32	>64
CAZ	0.03–64	154	64	>64
CZA	0.03–64	154	0.5	4
FEP	0.05–16	154	>16	>16
MEM	0.03–64	154	1	32
CIP	0.25–4	154	>4	>4
ATM	0.05–32	154	>32	>32
AMK	4–64	154	≤4	16
CST	0.5–8	154	≤0.5	1
TGC	0.25–4	154	≤0.25	1
<i>Escherichia coli</i>				
CFDC	0.03–64	42	0.06	0.5
CT	0.03–64	42	2	32
CAZ	0.03–64	42	4	64
CZA	0.03–64	42	0.25	0.5
FEP	0.05–16	42	8	>16
MEM	0.03–64	42	1	4
CIP	0.25–4	42	≤0.25	>4
ATM	0.05–32	42	32	>32
AMK	4–64	42	≤4	≤4
CST	0.5–8	42	≤0.5	1
TGC	0.25–4	42	≤0.25	≤0.25
<i>Klebsiella pneumoniae</i>				
CFDC	0.03–64	88	0.25	1
CT	0.03–64	88	64	>64
CAZ	0.03–64	88	64	>64
CZA	0.03–64	88	0.5	>64
FEP	0.05–16	88	>16	>16
MEM	0.03–64	88	1	64
CIP	0.25–4	88	>4	>4
ATM	0.05–32	88	>32	>32
AMK	4–64	88	≤4	>64
CST	0.5–8	88	≤0.5	1
TGC	0.25–4	88	0.5	1
<i>Enterobacter</i> sp.				
CFDC	0.03–64	24	1	4
CT	0.03–64	24	32	>64
CAZ	0.03–64	24	>64	>64
CZA	0.03–64	24	0.5	2
FEP	0.05–16	24	>16	>16
MEM	0.03–64	24	1	8
CIP	0.25–4	24	>4	>4
ATM	0.05–32	24	>32	>32
AMK	4–64	24	8	8
CST	0.5–8	24	≤0.5	1
TGC	0.25–4	24	1	1

^a MIC, minimum inhibitory concentration; MIC_{50/90}, MIC that inhibits 50% and 90% of the isolates, respectively

Table 5 In vitro activities of cefiderocol (CFDC), ceftolozane-tazobactam (CT), ceftazidime (CAZ), ceftazidime-avibactam (CZA), cefepime (FEP), meropenem (MEM), ciprofloxacin (CIP), aztreonam (ATM), amikacin (AMK), colistin (CST), and tigecycline (TGC) against Enterobacteriaceae (*E. coli*, *K. pneumoniae*, and *Enterobacter* sp.) producing NDM, VIM, or IMP carbapenemases

Organism group/antimicrobial agent	MIC range (mg/L)	No.	MIC ₅₀ ^a	MIC ₉₀ ^a
<i>Total Enterobacteriaceae</i>				
CFDC	0.03–64	134	1	4
CT	0.03–64	134	>64	>64
CAZ	0.03–64	134	>64	>64
CZA	0.03–64	134	>64	>64
FEP	0.05–16	134	>16	>16
MEM	0.03–64	134	32	>64
CIP	0.25–4	134	>4	>4
ATM	0.05–32	134	>32	>32
AMK	4–64	134	8	>64
CST	0.5–8	134	≤0.5	1
TGC	0.25–4	134	≤0.25	1
<i>Escherichia coli</i>				
CFDC	0.03–64	67	1	16
CT	0.03–64	67	>64	>64
CAZ	0.03–64	67	>64	>64
CZA	0.03–64	67	>64	>64
FEP	0.05–16	67	>16	>16
MEM	0.03–64	67	64	>64
CIP	0.25–4	67	>4	>4
ATM	0.05–32	67	>32	>32
AMK	4–64	67	8	>64
CST	0.5–8	67	≤0.5	1
TGC	0.25–4	67	≤0.25	≤0.25
<i>Klebsiella pneumoniae</i>				
CFDC	0.03–64	38	1	4
CT	0.03–64	38	>64	>64
CAZ	0.03–64	38	>64	>64
CZA	0.03–64	38	>64	>64
FEP	0.05–16	38	>16	>16
MEM	0.03–64	38	32	>64
CIP	0.25–4	38	>4	>4
ATM	0.05–32	38	>32	>32
AMK	4–64	38	16	>64
CST	0.5–8	38	≤0.5	1
TGC	0.25–4	38	1	2
<i>Enterobacter</i> sp.				
CFDC	0.03–64	29	1	4
CT	0.03–64	29	>64	>64
CAZ	0.03–64	29	>64	>64
CZA	0.03–64	29	>64	>64
FEP	0.05–16	29	>16	>16
MEM	0.03–64	29	16	64
CIP	0.25–4	29	1	>4
ATM	0.05–32	29	16	>32
AMK	4–64	29	≤4	>64
CST	0.5–8	29	≤0.5	≤0.5
TGC	0.25–4	29	0.5	1

^a MIC, minimum inhibitory concentration; MIC_{50/90}, MIC that inhibits 50% and 90% of the isolates, respectively

Table 6 In vitro activities of cefiderocol (CFDC), ceftolozane–tazobactam (CT), meropenem (MEM), ceftazidime (CAZ), ceftazidime–avibactam (CZA), colistin (CST), aztreonam (ATM), amikacin (AMK), ciprofloxacin (CIP), cefepime (FEP), and tigecycline (TGC) against *P. aeruginosa* producing a carbapenemase (either IMP, KPC, VIM, SPM, or GIM)

Organism group/antimicrobial agent	MIC range (mg/L)	No.	MIC ₅₀ ^a	MIC ₉₀ ^a
<i>Pseudomonas aeruginosa</i>				
CFDC	0.03–64	30	0.5	2
CT	0.03–64	30	>64	>64
CAZ	0.03–64	30	>64	>64
CZA	0.03–64	30	>64	>64
FEP	0.05–16	30	>16	>16
MEM	0.03–64	30	64	>64
CIP	0.25–4	30	>4	>4
ATM	0.05–32	30	8	64
AMK	4–64	30	8	>64
CST	0.5–8	30	≤0.5	1
TGC	0.25–4	30	8	>4

^a MIC, minimum inhibitory concentration; MIC_{50/90}, MIC that inhibits 50% and 90% of the isolates, respectively

≥8), and tigecycline (≤0.5/≤1). Elevated MIC₉₀ values for colistin were noted for 17 out of 101 strains for *K. pneumoniae* and 2 out of 14 strains for *Enterobacter* sp. For all other antibiotics, the MIC_{50/90} were superior or equal to the upper limit of the concentration range.

OXA-48-like producing Enterobacteriaceae were susceptible to a larger number of antibiotics compared to the KPC producers (Table 4). Again, cefiderocol is one of the antibiotics with the lowest MIC_{50/90} values, at 0.25 and 2 µg/mL, respectively. Direct competitors were ceftazidime–avibactam (0.5/4), meropenem (1/32), amikacin (≤4/16), colistin (≤0.5/1), and tigecycline (≤0.5/≤1). Again, for *Klebsiella* sp., the MIC₉₀ of CZA and MEM were affected by outliers and the MIC₅₀ values should be considered instead.

For the enterobacterial isolates producing NDM, VIM, or IMP carbapenemases (Table 5), the only antibiotics that had strong activity (MIC_{50/90}) were cefiderocol (1/4), colistin (≤0.5/≤1), and tigecycline (≤0.25/≤1).

Colistin-resistant strains, mainly Enterobacteriaceae, had high susceptibility to cefiderocol (≤0.5/≤2) and some activity for ceftolozane–tazobactam in the case of *E. coli* (0.25/>64), ceftazidime–avibactam (0.5/>64), meropenem (0.12/64), amikacin (≤4/16), and tigecycline (≤1/≤1). Except for cefiderocol and tigecycline, the MIC₉₀ values were close to or above the upper limit of the concentration range of the tested antibiotics for the Enterobacteriaceae being resistant to colistin. This could be explained by additional resistance traits, such as expression of genes encoding carbapenemases and ESBLs, in particular for *K. pneumoniae* and *Enterobacter* sp. The level of resistance to colistin (MIC₉₀ = 2) and of meropenem (MIC₉₀ = 1) was lower for *E. coli* than that noted for *K. pneumoniae* and *Enterobacter* sp.

Carbapenemase-producing *P. aeruginosa* were susceptible only to cefiderocol (0.5/2) and colistin (≤0.5/1) (Table 6). The same resistance trend was observed for carbapenemase-producing *A. baumannii* strains [CFDC (0.12/4) and CST

Table 7 In vitro activities of cefiderocol (CFDC), ceftolozane–tazobactam (CT), meropenem (MEM), ceftazidime (CAZ), ceftazidime–avibactam (CZA), colistin (CST), aztreonam (ATM), amikacin (AMK), ciprofloxacin (CIP), cefepime (FEP), and tigecycline (TGC) against *A. baumannii* producing an OXA-type carbapenemase (either OXA-23, -40, -58, or -72)

Organism group/antimicrobial agent	MIC range (mg/L)	No.	MIC ₅₀ ^a	MIC ₉₀ ^a
<i>Acinetobacter baumannii</i>				
CFDC	0.03–64	85	0.12	4
CT	0.03–64	85	16	>64
CAZ	0.03–64	85	>64	>64
CZA	0.03–64	85	16	64
FEP	0.05–16	85	>16	>16
MEM	0.03–64	85	32	64
CIP	0.25–4	85	>4	>4
ATM	0.05–32	85	>32	>32
AMK	4–64	85	64	>64
CST	0.5–8	85	0.5	1
TGC	0.25–4	85	1	2

^a MIC, minimum inhibitory concentration; MIC_{50/90}, MIC that inhibits 50% and 90% of the isolates, respectively

(0.5/1)], except that they were also susceptible to tigecycline (1/2) (Table 7). The only unexpected result is the overall low activity of ceftolozane–tazobactam against those *P. aeruginosa* isolates.

Among the 753 isolates tested, only 24 isolates exhibited an MIC value of cefiderocol ≥ 8 $\mu\text{g/mL}$, among which 45% were NDM producers ($n = 11$), 30% were OXA-23-producing *A. baumannii* ($n = 7$), and two VEB-, one SHV-, one VIM-, and one OXA-48-like producers among *K. pneumoniae*, *P. aeruginosa*, and *Enterobacter* sp. Noteworthy, cefiderocol was active against 68 out of 79 NDM producers, while most of the NDM producers co-produced other β -lactam resistance mechanisms, such as ESBLs (mostly CTX-M-15), porin defect, plasmid-mediated cephalosporinases, and other carbapenemases (data not shown). Cefiderocol was more active (MIC_{90} 2–4 mg/L) than the comparators (MIC_{90} >4 to >64 mg/L) (cephalosporins, carbapenem, fluoroquinolone, and monobactam) against all the tested strains. The only comparators with equal activity were colistin and tigecycline, with the limitation that tigecycline was not active against *P. aeruginosa*. Finally, it should be emphasized that cefiderocol displays much favorable pharmacokinetic parameters (tissue diffusion and use in renal impairment) than colistin and tigecycline [19], which will be an important factor for choosing adequate therapy of infections due to multidrug infections.

Compliance with ethical standards

Funding This work was funded by grants from the University of Fribourg and from Shionogi & Co., Ltd.

Conflict of interest None to declare.

Ethical approval Not applicable.

Informed consent Not applicable.

References

- Pitout JDD, Laupland KB (2008) Extended-spectrum β -lactamase-producing Enterobacteriaceae: an emerging public-health concern. *Lancet Infect Dis* 8(3):159–166
- Knothe H, Shah P, Krcmery V, Antal M, Mitsuhashi S (1983) Transferable resistance to cefotaxime, cefoxitin, cefamandole and cefuroxime in clinical isolates of *Klebsiella pneumoniae* and *Serratia marcescens*. *Infection* 11(6):315–317
- Philippon A, Labia R, Jacoby G (1989) Extended-spectrum β -lactamases. *Antimicrob Agents Chemother* 33(8):1131–1136
- Livermore DM (2003) Bacterial resistance: origins, epidemiology, and impact. *Clin Infect Dis* 36:S11–S23
- Gupta N, Limbago BM, Patel JB, Kallen AJ (2011) Carbapenem-resistant Enterobacteriaceae: epidemiology and prevention. *Clin Infect Dis* 53(1):60–67
- Kanj SS, Kanafani ZA (2011) Current concepts in antimicrobial therapy against resistant gram-negative organisms: extended-spectrum β -lactamase-producing Enterobacteriaceae, carbapenem-resistant Enterobacteriaceae, and multidrug-resistant *Pseudomonas aeruginosa*. *Mayo Clin Proc* 86(3):250–259
- Poirel L, Kieffer N, Liassine N, Thanh D, Nordmann P (2016) Plasmid-mediated carbapenem and colistin resistance in a clinical isolate of *Escherichia coli*. *Lancet Infect Dis* 16(3):281
- Liu YY, Wang Y, Walsh TR, Yi LX, Zhang R, Spencer J, Doi Y, Tian G, Dong B, Huang X, Yu LF, Gu D, Ren H, Chen X, Lv L, He D, Zhou H, Liang Z, Liu JH, Shen J (2016) Emergence of plasmid-mediated colistin resistance mechanism MCR-1 in animals and human beings in China: a microbiological and molecular biological study. *Lancet Infect Dis* 16(2):161–168
- Haenni M, Poirel L, Kieffer N, Châtre P, Saras E, Métayer V, Dumoulin R, Nordmann P, Madec JY (2016) Co-occurrence of extended spectrum β lactamase and MCR-1 encoding genes on plasmids. *Lancet Infect Dis* 16(3):281–282
- Hasman H, Hammerum AM, Hansen F, Hendriksen RS, Olesen B, Agersø Y, Zankari E, Leekitcharoenphon P, Stegger M, Kaas RS, Cavaco LM, Hansen DS, Aarestrup FM, Skov RL (2015) Detection of *mcr-1* encoding plasmid-mediated colistin-resistant *Escherichia coli* isolates from human bloodstream infection and imported chicken meat, Denmark 2015. *Euro Surveill* 20(49)
- Kohira N, West J, Ito A, Ito-Horiyama T, Nakamura R, Sato T, Rittenhouse S, Tsuji M, Yamano Y (2015) In vitro antimicrobial activity of a siderophore cephalosporin, S-649266, against Enterobacteriaceae clinical isolates, including carbapenem-resistant strains. *Antimicrob Agents Chemother* 60(2):729–734
- Ito A, Nishikawa T, Matsumoto S, Yoshizawa H, Sato T, Nakamura R, Tsuji M, Yamano Y (2016) Siderophore cephalosporin cefiderocol utilizes ferric iron transporter systems for antibacterial activity against *Pseudomonas aeruginosa*. *Antimicrob Agents Chemother* 60(12):7396–7401
- Tillotson GS (2016) Trojan horse antibiotics—a novel way to circumvent gram-negative bacterial resistance? *Infect Dis (Auckl)* 9: 45–52
- Ito-Horiyama T, Ishii Y, Ito A, Sato T, Nakamura R, Fukuhara N, Tsuji M, Yamano Y, Yamaguchi K, Tateda K (2016) Stability of novel siderophore cephalosporin S-649266 against clinically relevant carbapenemases. *Antimicrob Agents Chemother* 60(7):4384–4386
- Ito A, Kohira N, Bouchillon SK, West J, Rittenhouse S, Sader HS, Rhomberg PR, Jones RN, Yoshizawa H, Nakamura R, Tsuji M, Yamano Y (2016) In vitro antimicrobial activity of S-649266, a catechol-substituted siderophore cephalosporin, when tested against non-fermenting gram-negative bacteria. *J Antimicrob Chemother* 71(3):670–677
- Clinical and Laboratory Standards Institute (CLSI) (2015) Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically; Approved standards—tenth edition. CLSI document M07-A10. CLSI, Wayne, PA
- Huband MD, Ito A, Tsuji M, Sader HS, Fedler KA, Flamm RK (2017) Cefiderocol MIC quality control ranges in iron-depleted cation-adjusted Mueller–Hinton broth using a CLSI M23-A4 multi-laboratory study design. *Diagn Microbiol Infect Dis* 88(2): 198–200
- Clinical and Laboratory Standards Institute (CLSI) (2015) Performance standards for antimicrobial susceptibility testing; Twenty-fifth informational supplement. CLSI document M100-S25. CLSI, Wayne, PA
- Katsube T, Wajima T, Ishibashi T, Camilo Arjona Ferreira J, Echols R (2017) Pharmacokinetic/pharmacodynamic modelling and simulation of cefiderocol, a parenteral siderophore cephalosporin, for dose adjustment based on renal function. *Antimicrob Agents Chemother* 61(1). pii: e01381-16